Virtual Cell Version 4.0 Tutorial II

Creating a FRAP binding BioModel

Introduction

This tutorial is a continuation of the first tutorial. It is slightly more complicated in that in now includes reactive binding sites, unlabeled and labeled versions and bound and unbound versions of the nuclear protein RAN.

Following the Tutorial

You can create your own BioModel and Application as you read through the tutorial or you may choose to load the public version of this model. Go to View>Private Only and make sure this option is deselected so you will be able to view the public model in the Tutorial folder. Load the public BioModel “Tutorial_FRAPbinding”. There are two Applications, Compartmental and FRAPbinding, and the simulation results are saved as Simulation1 (for Compartmental) and Frap binding (for FRAPbinding).

Defining the biological model

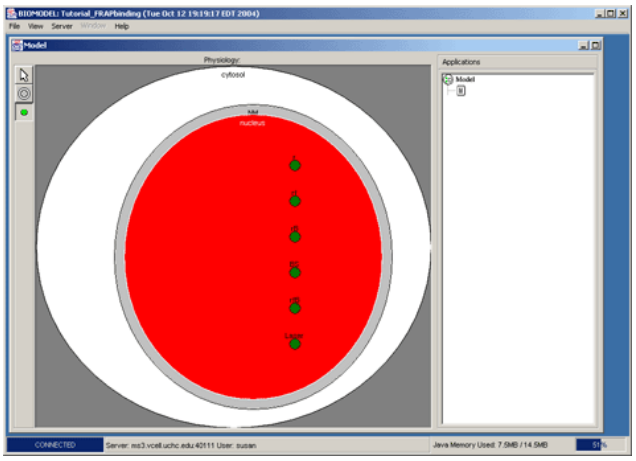
Creating and Defining Compartments

When the software starts, you are presented with an undefined BioModel. Select the compartment once with the left mouse button, and then use the right mouse button to access the Properties menu. Enter "cytosol" in the Feature Name text field and press OK.

Select the compartment tool  once and click in the cytosolic compartment. Type in "nucleus" in the Feature Name text field, and "NM" (Nuclear Membrane) in the Membrane Name text field; press Add Feature.

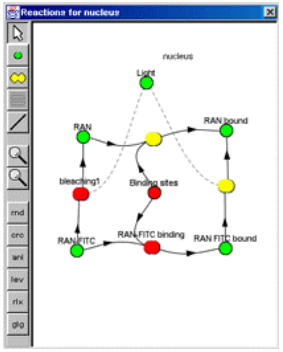
Adding Species

Select the species tool  and then click in the nuclear compartment once. In the Add New Species dialog, enter "r" in the Name text field. Press Annotate and type in “RAN” in the text field and press OK.
Continue to use the species tool to add the following Species, using the abbreviations listed in the table.



RAN-FITC	rf
RAN_bound	rB
Binding_sites	BS
RAN_FITC_bound	rfB
Light	Laser

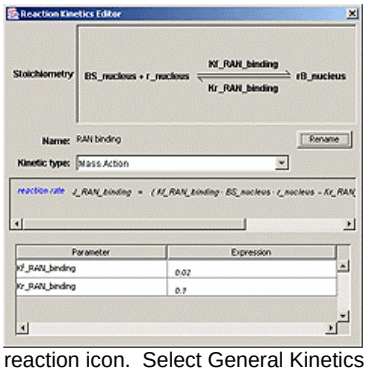
Your model should look similar to the picture when you have added all the species.
You may want to save the model before proceeding with the reactions. Go to File>Save As and enter a unique name in the text field, press Save.



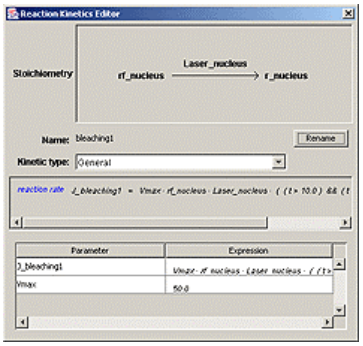
Defining Reactions

Select the nuclear compartment and then use the right mouse button to access the Reactions menu. Arrange the species in the Reactions dialog so that each is visible. You might want to use this image as a guide for organizing the reactions.

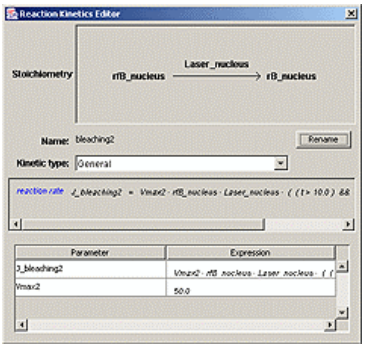
Select the reaction icon  and then click once in the nuclear compartment. Do this three more times. Arrange the reaction icons as in the picture above. Select the reaction icon, one at a time, and use the right mouse button to access the Properties option that will open the Reaction Kinetics Editor. Click the rename button in each editor and give the following reaction names:RAN_binding, bleaching 1, bleaching 2 , and RAN_FITC_binding



Use the line tool to connect the species to the reaction icons as in the image above. The reaction icon is set up such that the left side of the icons is for reactants and the right side is for products. You have to drag to the middle of the reaction icon with the Laser species. The laser is acting as a catalyst for the bleaching reactions.
Select the RAN_binding reaction icon and use the right mouse button to access the Properties option. Press Rename to name the reaction. This name will appear when the reaction icon is selected. Select Mass Action for the Kinetic Type from the selection list. Double click the Expression text field for Kf_RAN_binding, and enter ".02". Double click the Expression text field for Kr_RAN_binding and enter ".1". Close the Editor when you have finished.

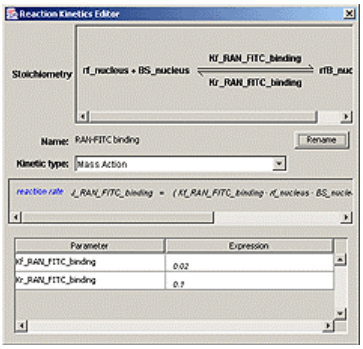


Open the Reaction Kinetics Editor for the bleaching 1 reaction icon. Select General Kinetics for the Kinetic type and enter the following flux reaction equation: (Vmax*rf_nucleus*Laser_nucleus*((t>10.0)&&(t<10.5))). Vmax will appear as a parameter. Double click the Expression text field and type in "50.0". Press Enter to accept the value. This equation defines the bleaching period as .5 seconds, starting after 10 seconds.



Open the Reaction Kinetics Editor for the bleaching 2 reaction icon. Select General for the Kinetic type and enter the following Rate Equation: (Vmax2*rfB_nucleus*Laser_nucleus*((t>10.0)&&(t<10.5))). Vmax2 will appear as a parameter. Double click the Expression text field and type in "50.0". Press Enter to accept the value. This equation defines the bleaching period as .5 seconds, starting after 10 seconds.

There are two separate bleaching reactions to account for bound and unbound RAN. Open the Reaction Kinetics Editor for the RAN_FITC_binding reaction icon. Select Mass Action for the Kinetic Type and enter ".02" for a Forward Rate, Kf, and ".1" for a Reverse Rate, Kr. Once all the reactions have been entered, save the model and proceed to Application.



The FRAP binding Application

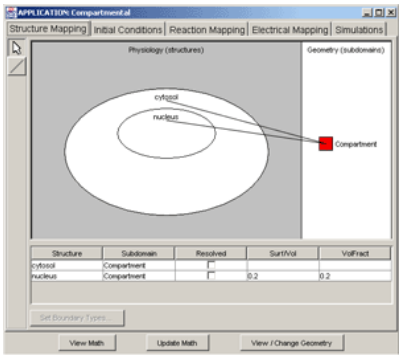
Introduction

Each model being developed requires an Application, which consists of a detailed description of the cellular geometry, Structure Mapping, Initial Conditions, and Reaction Mapping. The geometry represents the morphometry of a particular cell or portion of a cell. The geometry may be captured by various imaging modalities such as wide field, confocal, or electron microscopy. Analytic geometry may be used to define very regular structures or symmetric cells.

Creating the geometry

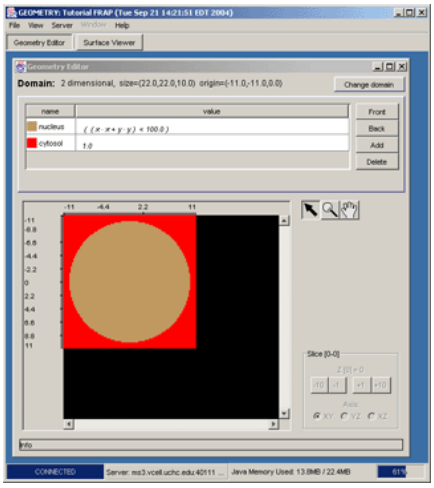
In this model you will initially create a Compartmental model where the BioModel is mapped to a single point. Run the simulation, and after you review your results create a spatial model. In the Spatial model you will map the BioModel to a defined Geometry and run another simulation. The results for the Compartmental model will give you an initial idea about how your model is performing. The results you get in the Spatial model, although they will be more accurate, should be similar to those obtained in the Compartmental model.

I. Compartmental Geometry

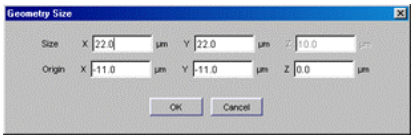


A Compartmental model represents a single point simulation based on the defined physiological model and the geometric assumptions, the Surface to Volume Ratio and Volume Fraction. Compartmental models are not spatially resolved. The compartmental models are solved using nonlinear ordinary differential equations. These equations are generally computed within seconds.
In the Application panel of the BioModel document go to Application>New and enter "compartmental" for the Application name. The Application will initialize with the Structure Mapping panel. The compartmental model, the default model, is automatically mapped to a single compartment.

II. Spatial Geometry



In the BioModel document, go to File>New>Geometry>Analytic> 2-D. You will be presented with the Geometry document which contains a single subVolume viewable in the Geometry Editor. Press Add one more time to create an additional subVolume. The two subVolumes that you create will represent the extracellular and cytosolic compartments. Select one of the subVolumes and double click the Name text field. Type in "cytosol" and press Enter. Double click the Value text field and enter the following equation to define a circle: ((x*x)+(y*y))<100.0 Press Enter after entering the equation.
Double click the Name text field for the other subVolume and enter "extracellular". Press the Change domain button to access the Geometry size dialog. Enter "22" in the X and Y size text fields, and enter "-11" in the X and Y origin text fields. Press OK to accept the values and to close the window.



If the circle you just defined is not visible, make sure to select the cytosolic volume and press Front to bring it in front of the extracellular volume. The Front and Back buttons set the positioning of the subVolumes. It is important to arrange the subVolumes in the list so they are calculated in the proper order. If a subVolume is hidden or unreachable, it will not be calculated.
Save the Geometry by going to File>Save.

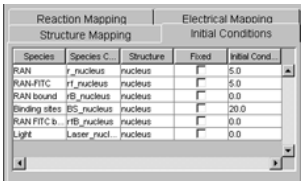
Creating the Applications

I. Compartmental Application

Structure Mapping

On the Structure Mapping tab, double click the SurfVol column for the nucleus. Type in ".2" for the Surface to Volume Ratio and press Enter to accept the value.

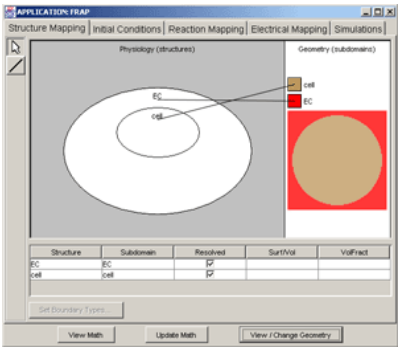
Initial Conditions



Select the Initial Conditions tab. Double click in the Initial Conditions text field for RAN and enter an initial concentration of 5.0. Press OK to accept the value and to dismiss the dialog. Do the same for RAN-FITC, enter a value of 5.0 and for Binding sites enter a value of 20.0.
You may want to resave your model at this point and proceed to Compartmental Simulation.

II. Spatial Application

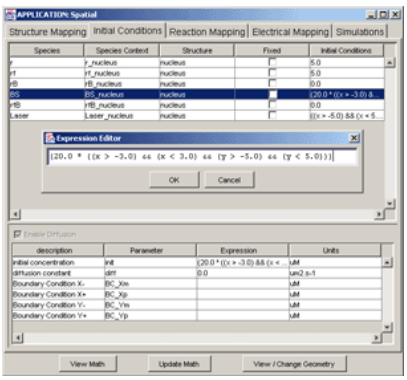
Structure Mapping



Return to the BioModel document, and in the Application panel, go to Application>New. Press View/Change Geometry. Select the geometry you just created from the Geometry database. The geometry will appear in the Structure Mapping tab of the Application dialog.

Use the line tool  to map the physiology model to the geometric representation. Map extracellular to extracellular and cytosol to cytosol. You need to reselect the line tool each time you do a mapping, and you need to map from the physiology to the geometry.

Initial Conditions



Select the Initial Conditions tab. Double click the Initial Conditions text field for RAN (r_nucleus), enter 5.0 in the Expression Editor and enter 10 in the Diffusion Rate text field. Remember to press Set for the Diffusion Rate.
Enter the same values, in the same manner, for RAN-FITC (rf_nucleus) as you did for RAN; 5.0 for an Initial Concentration and 10.0 for the Diffusion Rate.
Select Binding_sites (BS_nucleus). Double click the Initial Conditions text field and enter the following equation in the Expression Editor text box: (20.0*((x>-3.0)&&(x<3.0)&&(y>-5.0)&&(y<5.0)))
Double click the Initial Conditions text field for the Laser. Type in the following equation and press OK to accept the equation: ((x>-5.0)&&(x<5.0)&&(y>-5.0)&&(y<5.0))

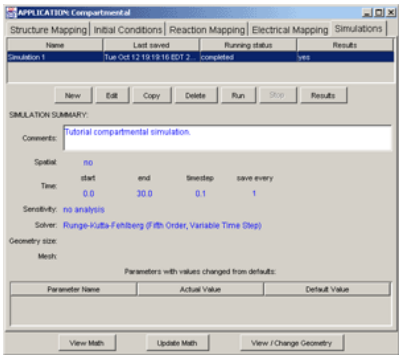
Reaction Mapping

Leave the Reaction Mapping as the default. The reactions should all be enabled. Fast

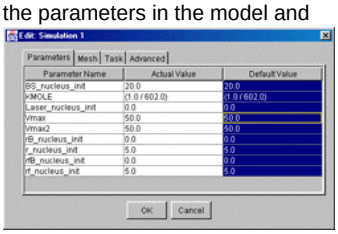
kinetics should not be enabled. Resave your model and proceed to Spatial Simulation.

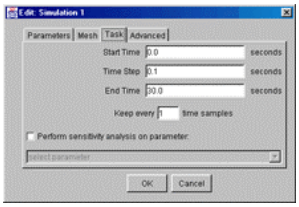
Running the FRAP Binding Compartmental and Spatial Simulations

I Compartmental Simulation and Results

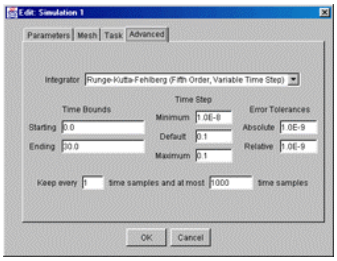


Select the Simulation tab and press the New button. In the upper panel of the Simulations dialog, a simulation with a default name will appear. Double click the Name text field and type in a simulation name. You can add additional notes regarding the simulation in the Comments text field if you choose to do so.
Press the Edit button to access the Parameters tab that lists all the parameters in the model and their corresponding values, Actual and Default. You can change the values and run new simulations without having to rebuild a new model. Double click the Actual Value text field for a Parameter and enter a new value. Altered values appear in red text. In this tutorial we will not alter any values. Press the Task tab to define the run conditions for the simulation, as listed in the table.

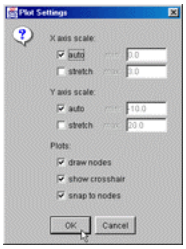


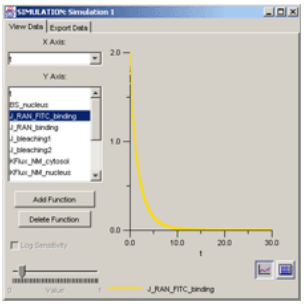


Start Time: 0
Time Step: 0.1
End time: 30.0
Save Interval: 1



The Advanced tab has various integrators to choose from. Use all but one of the default settings for this example. The only change you will make is to the Maximum Time Step. Change this from the default of 1.0 to 0.1.
Make sure your simulation is still selected when you press the Run button to initiate the simulation. Your model will automatically be resaved with the new run conditions and the simulation will begin. The results are stored on the remote database server.
Once the simulation has generated some results, the Status field will display Complete and the Results field will display Yes. The simulation must be selected for the Results button to be active. Press the Results button to open the Results dialog or wait until the simulation is complete and the dialog will open automatically.
You can select how you want the graph constructed by choosing the parameters for the X and Y axes. The graph is interactive; put your mouse over the graph to see the coordinates for each data point on the curve. Press the right mouse button to access the Plot setting dialog for additional graphing options.

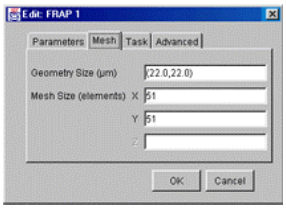




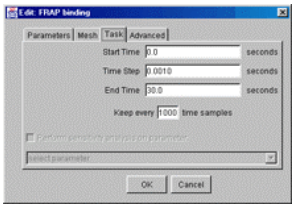
Export Features

Press the Export Data tab on the Simulations dialog. Results from compartmental simulations can be exported as Comma Delimited ASCII files which can be opened in graphing programs such as Excel. See Chapter 9 of the user guide, Exporting Simulation Results, for more information.

II Spatial Simulation and Results



Press the Simulations tab on the Application dialog and press the New button. Double click in the Name text field to name the simulation.
Press the Edit button to access the Mesh and Task tabs. Select the Mesh tab, enter "51" for the X and Y dimensions for the Mesh Size. The Geometry Size, which was defined in the Geometry Editor, should be listed as (22.0, 22.0, 0).



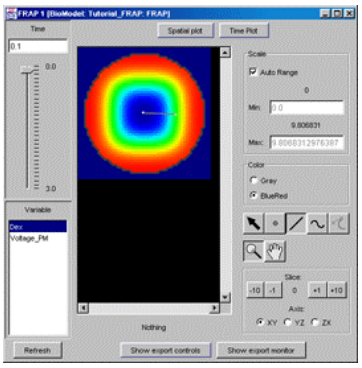
Press the Task tab to define the run conditions for the simulation, as described in the table. Press OK to accept the conditions and to close the dialog.

Start Time: 0

Time Step: 0.001

End Time: 30.0

Save Interval: 1000

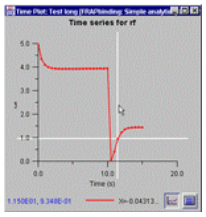


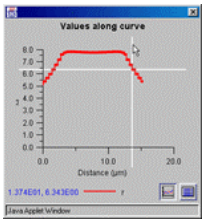
Make sure your simulation is still selected when you press Run to initiate the simulation. Your model will automatically be resaved with the run conditions and the simulation will begin. The results from the simulation are automatically stored on the remote database server.

Once results have been generated, press the Results button to access the Results dialog. The simulation must be selected to activate the Results button.

Use the scroll bar, on the left side of the Results dialog, to change the time interval or enter a time in the Time text field and press Enter. You can drag the scroll bar or select it and then use the up and down arrows on your keyboard to step through the time points.

You can display your results in either a Gray or a Blue-Red color map. You may toggle between auto and manual scaling. Enter values in the Min and Max text fields for manual scaling. Remember to press Enter to accept the value and to update the image display.

Use the Point tool to generate a Time Plot, and the Line and Spline tools to generate a Spatial Plot. You can choose between displaying your results as a plot or viewing the data values. Press your right mouse button, while over the 



graph, to access the Plot Settings dialog.

Export Features

Press Show export controls to display the export features. Select the variable(s), time interval and data region(s) you wish to export. Files may be exported as Comma delimited ASCII files, QuickTime move files, GIF89a image files or Animated GIF files. See Chapter 10 of the user guide, Exporting Simulation Results, for more information.